changes relative to the previous version, the changes indicated are relative to the originally filed version.

The specification has also been amended to correct an inadvertently made, obvious error in the first entry in Table 1 to indicate that Oligo #164 (SEQ ID NO:1) is an antisense oligonucleotide. This error is obvious from the fact that the hybrid, inverted hybrid, and inverted chimeric oligonucleotides are based on this same sequence and the control oligonucleotides are mismatched from this sequence.

Additionally, the specification has also been amended to delete inadvertently inserted data obviously unrelated to this application and to correct the heading of the section containing this data to reflect the data contained therein. Support for the amendment of the heading can be found in one of the parent patents, U.S. Patent Number 5, 969, 117, at Col. 12, line 11, which used the same heading.

Finally, the specification has also been amended to correct the description of Example 10. These changes are obvious from a comparison of Table 1, amended herein, which shows the sequences and SEQ ID NOS, with Figure 1, which shows the results of the experiment.

Thus, no new matter has been introduced by these amendments.

Applicant assumes that all rejections not repeated in the Office Action of June 20, 2001 have been overcome and are withdrawn.

Applicant acknowledges that the formal drawings have been approved by the Draftsperson.

The outstanding rejections are addressed individually below.

1. ***Claims 1-33 are enabled by the specification as filed.***

Claims 1-33 stand rejected under 35 U.S.C. § 112, first paragraph, as not being enabled by the specification as filed. Applicants point out that claims 21 and 22 were cancelled in the response filed on March 26, 2001, and thus the rejection of claims 21 and 22 is moot. Applicant respectfully traverses the rejection of claims 1-20 and 23-33.

Claim 1 is directed to a method for inhibiting proliferation of cancer cells comprising administering to the cells a first agent comprising a synthetic, modified oligonucleotide complementary to, and which down-regulates the expression of, nucleic acid encoding protein kinase A subunit RIα, the modified oligonucleotide having from about 15 to about 30 nucleotides and being a hybrid, inverted hybrid, or inverted chimeric oligonucleotide of specific characteristics, administering to the cells a second agent comprising an antibody that binds to EGFR or a chemotoxic agent selected from an enumerated group, wherein the administering steps may be performed simultaneously or sequentially in any order. Other independent claims are further directed to a pharmaceutical composition and a method for treating cancer.

The Office Action states that the specification, while being enabling for inhibiting proliferation of cancer cells *in vitro*, does not reasonably provide enablement for treatment of cancer in a patient *in vivo*, and that the specification does not enable a person skilled in the art to which it pertains to use the invention commensurate in scope with these claims. Applicant maintains that the specification enables the pending claims.

The Office Action further states that the instant application does not provide sufficient enablement for one of skill in the art to practice the full scope of the claimed invention without undue experimentation. (Office Action, page 3). The Office Action cites Crooke (*Antisense Research and Application*, Chapter 1, Basic Principles of Antisense Therapeutics, Springer-Verlag, Berlin, Heidelberg, New York, page 3, 1998) for a variety of factors that influence cellular uptake and distribution of antisense based therapeutics, and states that due to the unpredictability in cellular behavior associated with variations in sequence, length, and modifications of the oligonucleotides encompassed by the present invention, it is likely that the examples comprising the use of the HYB 165 oligonucleotide are not representative of all oligonucleotides encompassed by the claimed invention. (Office Action, page 4) Applicants respectfully disagree.

M.P.E.P § 2164.01 states that 35 U.S.C. § 112, first paragraph, "has been interpreted to require that the claimed invention be enabled so that any person skilled

in the art can make and use the invention without undue experimentation." The same section further states that "[t]he fact that experimentation may be complex does not necessarily make it undue, if the art typically engages in such experimentation." M.P.E.P § 2164.02 states that "[a]n *in vitro* or *in vivo* animal model example in the specification, in effect, constitutes a 'working example' if that example 'correlates' with a disclosed or claimed method invention. . . . In this regard, the issue of 'correlation' is also dependent on the state of the prior art. In other words, if the art is such that a particular model is recognized as correlating to a specific condition, then it should be accepted as correlating unless the examiner has evidence that the model does not correlate." This section further states that a "rigorous or an invariable exact correlation is not required . . . ." M.P.E.P § 2164.03 relates to the relationship of predictability of the art and the enablement requirement; this section states that "what is known in the art provides evidence as to the question of predictability."

Applicant submits that one of ordinary skill in the art would know how to determine effective antisense oligonucleotides without undue experimentation. For example, Milner et al. (*Nature Biotechnology* (1997) **15**:537-541; attached hereto as Appendix A) demonstrates "a combinatorial technique that allows simultaneous assessment of all possible ONs [oligonucleotides] within a given region identifying sequences open to duplex formation. An oligonucleotide 'scanning' array reduces the number of synthesis steps while providing a parallel and exhaustive analysis of all ONs in the region to be targeted." (page 537) This article further states that "those ONs which give high duplex yield on the array proved to be effective antisense agents in in vitro RNase H and translation studies." (page 537) As stated in the abstract, "the arrays provide a simple empirical method of selecting effective antisense oligonucleotides for any RNA target of known sequence." Thus, based on public information, Applicant submits that based on this information, one of ordinary skill in the art would be able to determine effective antisense oligonucleotides without undue experimentation.

Milner et al. also state that "hetroduplex yield on the array correlated well with in vivo and in vitro cell culture antisense activities." (page 540) Milner et al. discusses a reference by Monia et al. (*Nature Medicine* (1996) **2**:668-675; attached hereto as Appendix B), which identified an antisense inhibitor, ISIS 5132. (see page 669) ISIS 5132 was found to display "very potent inhibitory effects" *in vivo* (page 671) and was one of the antisense inhibitors that inhibited expression of C-*raf* in cell culture and *in vivo*. (page 672) This antisense inhibitor was also found to show *in vivo* antitumor effects against two additional tumor cell lines. (page 672) Milner et al. conducted a blind experiment, performing analysis on a scanning array that picked out ISIS 5132 as one of two high-yielding oligonucleotides in a 100 b region around the oligonucleotide. (page 540) Furthermore, as discussed in detail below, many published articles indicate that antisense oligonucleotides have been shown to be effective; therefore, the examples in the specification showing enablement of the invention with respect to one oligonucleotide *in vivo* should be enabling for the other oligonucleotides encompassed by the claims.

Evidence from Galderisi et al. (*J. Cell. Physiol.* (1999) **181**:251-57; attached hereto as Appendix C), indicates that intravenous administration of phosphorothioate oligodeoxynucleotides showed effective and specific antisense inhibition in animal models, that antisense oligodeoxynucleotides have been shown to be effective in preclinical studies, and that some antisense oligodeoxynucleotides have reached clinical trials. The article also teaches that a drug based on antisense technology is now available in the United States. This article provides antisense examples suggesting that such compounds have some therapeutic efficacy, including their use as antiviral agents.

In addition, Agrawal states, at page v of Antisense Therapeutics, (Sudhir Agrawal, ed.) 1996, (cited pages of which are attached hereto as Appendix D), that "[t]he results of preclinical studies using oligodeoxynucleotide phosphorothioates have shown that antisense oligonucleotides have good biological activity, pharmacology, pharmacokinetics, and safety both in vitro and in vivo, and they are currently being

evaluated in human clinical trials for the treatment of viral infections and cancers." Zamecnik (also Appendix D) states at page 6 of the same book that the synthetic antisense oligonucleotide technology displays promising results in cell-free systems, tissue cultures, and animal models and is at early trial points in human testing against HIV, leukemia, Herpes virus, and other diseases.

Craig et al. (*Exp. Opin. Ther. Patents* (1997) 7:1175-1182; attached hereto as Appendix E) teaches at page 1177 that once a modification to the oligonucleotide backbone "is found to confer a favorable characteristic, it can then be used in oligonucleotides having different sequences of nucleosides and, thus, provide utility for the treatment of other diseases" as well as discussing information regarding the patentability of antisense technology.

Furthermore, Monia et al., discussed above, states at page 668 that the authors demonstrate that "treatment of human tumor cells with appropriate phosphorothioate antisense ODNs leads to specific inhibition of *C-raf* gene expression in cell culture and *in vivo*. Moreover, antisense-mediated inhibition of *C-raf* gene expression in human tumor cells results in potent antiproliferative effects in cell culture and potent antitumor activity *in vivo*." As discussed above, this article identified an antisense inhibitor, ISIS 5132, (see page 669), which was found to display "very potent inhibitory effects" *in vivo*. (page 671) This antisense inhibitor was found to show *in vivo* antitumor effects against two additional tumor cell lines. (page 672)

As stated in the Amendment filed on March 26, 2001, Examples 27, 28, and 29 (pages 90-95) as well as Figures 16, 17, and 18 of the instant patent application provide examples and data indicating that the claimed invention does work *in vivo* in an accepted animal model. More specifically, Example 27 indicates that HYB 165 inhibits tumor growth after intraperitoneal or oral administration in mice. The data for this experiment is presented in Figures 16A and 16B. Example 28 indicates that oral HYB 165 cooperatively inhibits tumor growth and increases survival in combination with taxol. Data for this experiment is presented in Figures 17A and 17B. Example 29 indicates that the cooperative antitumor effect of HYB 165 with taxol is accompanied by

inhibition of new vessel formation and growth factor production as well as other results of histochemical analysis. Data for this experiment is presented in the table in Figure 18. Additional support for the *in vivo* use of the methods and pharmaceutical compositions of the invention is found in the description of the figures in the specification at page 20, lines 9-29. Further support for the preferred dosages for the cytotoxic agents and oligonucleotides is found in the specification at page 27, line 15 to page 28, line 14 and page 29, line 8 to page 30, line 10.

Additionally, Example 10 indicates that a single dose of RI_{α} antisense, hybrid, or inverted hybrid oligonucleotide was tested by injection into the right flank of athymic mice previously innoculated with tumor cells and tumor volumes were obtained. (page 58, line 25 to page 59, line 20), and the results are shown in Figure 1. Thus, it is not only HYB 165 that was shown to work *in vivo*.

The specification also indicates that *in vitro* experiments were performed analyzing, *inter alia*, the effect of inverted hybrid or inverted chimeric structure on oligonucleotide-mediated mitogenicity (page 50, line 16 to page 52, line 6) and to determine the ability of inverted hybrid oligonucleotides and inverted chimeric oligonucleotides to activate RNase H *in vitro* when bound to a complementary RNA molecule (page 56, line 4 to page 57, line 28).

These teachings clearly indicate that the specification enables the claimed invention for both *in vitro* and *in vivo* use by providing supportive data indicating that *in vivo* use of the invention has, in fact, been achieved.

Based on the information provided in the published references described above, the data in the specification indicating that HYB 165 (SEQ ID NO:4) and other oligonucleotide sequences were shown to be operable *in vivo*, and the data showing that other oligonucleotides were operable *in vitro*, Applicant submits that: (1) at least one oligonucleotide works *in vivo* (and in fact more than one oligonucleotide has been shown to work *in vivo*); (2) because there is a correlation between *in vitro* and *in vivo* results, there is a reasonable expectation that antisense oligonucleotides shown to work

in vitro would also be expected to work *in vivo*; (3) it would not require undue experimentation to find other oligonucleotides that would be functional besides HYB 165; and (4) claims only cover operable embodiments, and as stated in M.P.E.P § 2164.03 "even in unpredictable arts [Applicant submits that this art is no longer unpredictable], a disclosure of every operable species is not required." Therefore, applicant submits that the specification enables the scope of the claimed invention.

The Office Action further states that the instant claims read on a method wherein the oligonucleotide of the invention "consists essentially of the nucleotide sequence set forth in SEQ ID NO: 4," and that it is unclear what other sequences are encompassed by this language. Applicant notes that only claims 3, 14, and 24 include such language. Applicant also notes that this language was used in issued claims in a parent application, U.S. Patent No. 5,969,117 (attached hereto as Appendix F). Applicant submits that this language is clear because it indicates that the nucleotide sequence is that sequence set forth in SEQ ID NO: 4.

Therefore, Applicant submits that in view of the foregoing remarks and the references submitted, pending claims 1-20 and 23-33 are enabled by the specification as filed. Accordingly, Applicant respectfully requests that the rejection of these claims under 35 U.S.C. § 112, first paragraph, be reconsidered and withdrawn.

CONCLUSIONS

In view of the arguments set forth above, Applicant respectfully submits that the rejections contained in the final Office Action mailed on June 20, 2001, have been overcome, and that the claims are in condition for allowance. If the Examiner believes that any further discussion of this communication would be helpful, she is invited to contact the undersigned at the telephone number provided below.

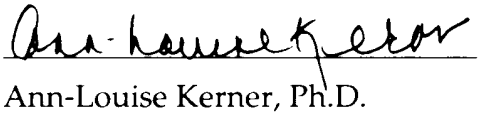
Applicant encloses herewith a Petition for a One Month Extension of Time pursuant to 37 C.F.R. § 1.136, to respond to the Examiner's Office Action mailed on June

20, 2001. Our deposit account no. 08-0219 is to be charged the $55.00 fee for this purpose.

Applicant also encloses herewith a Supplemental Information Disclosure Statement. Please charge Deposit Account No. 08-0219 the $180.00 fee for this submission.

No other fees are believed to be due in connection with this response. However, please charge any underpayments or credit any overpayments to Deposit Account No. 08-0219.

Respectfully submitted,



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Marked Up Version of Replacement Paragraph in Specification Under 37 C.F.R. §1.121 (b)(1)(iii) Compared to Original Version

Cross-Reference to Related Applications:

This application is a ~~non-provisional~~ continuation-in-part application claiming priority from U.S.S.N. 60/103,098, filed on October 5, 1998, and from U.S.S.N. 09/022,965, filed on February 12, 1998, which is a continuation-in-part application of U.S.S.N. 08/532,979, filed September 22, 1995**, which issued as U.S. Patent No. 5,969,117, which is a continuation-in-part application of U.S.S.N. 08/516,454 filed August 17, 1995, which issued as U.S. Patent No. 5,652,356**.

First entry in Table 1 at page 22:

164	GCG TGC CTC CTC ACT GGC ~~Control~~ **Antisense**	1

Heading at page 36, lines 26-27:

~~Propagation and Quantitation of Cell Lines and Virus Stocks~~

In Vitro Complement Activation Studies

Paragraph at page 58, line 25 to page 59, line 20:

LS-174T human colon carcinoma cells (1×10^6 cells) were inoculated subcutaneously (s.c.) into the left flank of athymic mice. A single dose of RI_α antisense hybrid (Oligo ~~164~~ **165**, SEQ ID NO:4), inverted hybrid (Oligo 166, SEQ ID NO:6), or ~~inverted chimeric~~ **antisense** (Oligo ~~190~~ **164**, SEQ ID NO:1) oligonucleotides or control oligonucleotide (Oligo 169, SEQ ID NO:7); Oligo 168 (SEQ ID NO:5); Oligo 188, (SEQ ID NO:3)~~)~~ as shown in Table 1 (1 mg

per 0.1 ml saline per mouse), or saline (0.1 ml per mouse), was injected s.c. into the right flank of mice when tumor size reached 80 to 100 mg, about 1 week after cell inoculation. Tumor volumes were obtained from daily measurement of the longest and shortest diameters and calculation by the formula, $4/3\pi r^3$ where r = (length + width)/4. At each indicated time, two animals from the control and antisense-treated groups were killed, and tumors were removed and weighed. The results are shown in FIG. 1. These results show that the size of the tumor in the animal treated with the inverted hybrid oligonucleotide 166 having SEQ ID NO:6 was surprisingly smaller from three days after injection onward than the phosphorothioate oligonucleotide 164 having SEQ ID NO:1. That this effect was sequence-specific is also demonstrated in FIG. 1: control oligonucleotide 168 (SEQ ID NO:~~3~~ **5**) has little ability to keep tumor size at a minimum relative to the hybrid and inverted hybrid oligonucleotides.